NOTES ON THE LIFE-CYCLE OF AZYGIA ACUMINATA GOLDBERGER, 1911 (AZYGIIDAE-TREMATODA)

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Studies of trematodes belonging to the genus *Azygia* Looss, 1899 from North American fresh-water fish, date from the work of Leidy (1851). He described *Distoma longum* from the stomach of *Esox ester* LeSueur, 1818 from near Cleveland, Ohio. Since that time many workers have added to knowledge of the North American species belonging to this genus. Manter (1926) gave a systematic review of the family *Azygiidae* and stated (p. 57) that "Azygia is the only genus of the family showing taxonomic confusion in its species." He further pointed out that these forms are all very muscular and highly contractile, which not only alters the general shape of the worm but also changes the relative position of such structures as the acetabulum and the reproductive organs. He also mentioned that size of the eggs and distribution of the vitellaria vary considerably within a species and cannot be relied on as taxonomic characters. Manter recognized three valid North American species: *A. acuminata* Goldberger, 1911, *A. angusticauda* (Stafford, 1904) and *A. longa* (Leidy, 1851).

Van Cleave and Mueller (1934) endorsed the action of Manter in reducing the number of species in North America and felt that *A. acuminata* should also be reduced to synonymy with *A. longa*.

Stunkard (1956) gave a chronological account of the genus *Azygia* and noted (p. 266) "—discordant observations and divergent opinions," concerning the proposed specific and generic names for members of the genus. This thorough account need not be repeated here. Stunkard recognized *A. sebago* as a distinct species and suggested that the European *A. lucii* may also be present in North America and that it possibly is distinct from *A. longa*.

None of the species have been studied to determine the extent of variation that normally occurs as the result of early development in a wide variety of paratenic hosts (small fishes and planarians) and further development in varied definitive hosts (large fishes). Until such studies are undertaken, the taxonomic picture of the group will remain confused.

It is evident, in reviewing the literature, that species proposed by various North American authors and since placed in synonymy may indeed be valid species. In such worms, which are very muscular and highly contractile, size and shape can not be relied on as taxonomic characters except in a very general way. The fact that these worms become sexually mature while relatively small and continue to grow throughout life further complicates the taxonomic picture. *A. angusticauda* is the only North American species which can be readily distinguished from the other species described from this continent. It can be separated because of the more

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posterior position of the acetabulum and the presence of the gonads in the posterior one-sixth of the body (usually within the posterior one-seventh to one-eighth of the body). The rest of the species comprise a complex composed of *A. longa, A. sebago, A. lucii* and *A. acuminata.* This complex may perhaps include as yet unrecognized species, or species which at the present time are represented as identical to *A. longa.*

The descriptions of the cercaria of *A. longa* by Sillman (1953a, 1953b), of the cercaria of *A. sebago* by Stunkard (1956), and the description of the cercaria of *A. acuminata* in the present report complete the life-histories of all of the recognized species of *Azygia* in North America with the possible exception of *A. lucii* and *A. angusticauda.* Dickerman (1937) named *Cercaria angusticauda* as a new species, but did not describe the larva.

Moreover, in discussing the cystocercous cercariae of North America Horsfall (1934) pointed out that *C. wrighti* Ward, 1916, *C. anchoroides* Ward, 1916, and *C. brookoveri* Faust, 1918 appear to be typically azygiid in morphology and should logically develop into adult azygiids. One of these cercariae may develop into *A. lucii,* but if so it is morphologically distinct from the European larva.

Szidat (1932) showed that *Cercaria mirabilis* Braun is the larva of *A. lucii* in Europe. Sillman (1953a) described the larva of *A. longa* from the snail, *Amnicola limosa,* on the basis of experimental and limited natural infections. Sillman (1953b) added additional information concerning the life-cycle of *A. longa* which was reported from both the mud pickerel, *Esox vermiculatus* and *Amia calva* in the vicinity of Ann Arbor, Michigan. Sillman also assigned worms from the bowfin, *Amia calva,* to the suppressed species, *A. acuminata.* He was unable to experimentally obtain infections of *Amnicola* from eggs of *A. acuminata.*

Stunkard (1950) identified larval distomes from the pharyngeal pockets of planarians, *Dugesia tigrinum,* as immature azygiids. Planarians serve as paratenic hosts since further development of the distomes does not occur in this host. Stunkard (1956) reported on the life-cycle of *Azygia sebago* and experimentally showed that larval stages develop in *Amnicola limosa;* further that the cystocercous cercaria is distinct from that reported by Sillman (1953a, 1953b) from the same snail host. Stunkard also reported that these cercariae were ingested by small fish (guppies and small blue gill sunfish, *Lepomis macrochirus*) and also were obtained experimentally and naturally from the pharyngeal pockets of planarians, *Dugesia tigrinum.* He stated (p. 265), "How the larvae reach the pharyngeal pockets is not clear." On the basis of known feeding habits of planarians, he correctly suggested the manner in which the distomes enter the pharyngeal pockets but did not observe it.

In preliminary observations during the present study, cystocercous cercariae belonging to both *A. sebago* from *Amnicola limosa* and *A. acuminata* from *Campeloma decisum* (Say) were placed in finger bowls with *Dugesia tigrinum.* The planarians reacted to close proximity of both species of larvae by raising the anterior end of the body and, when the swimming cercaria came in contact with the under surface of the planarian, actively enclosing the cercaria against the glass container. The pharynx was then extruded and the cercaria was sucked into the protruded pharynx. Usually the tail entered first, always so in cercaria of *A. acuminata.* The distome portion of *A. sebago* was sucked into the pharynx where it was gradually divested of the tail and the distome became active and eventually

PLATE I

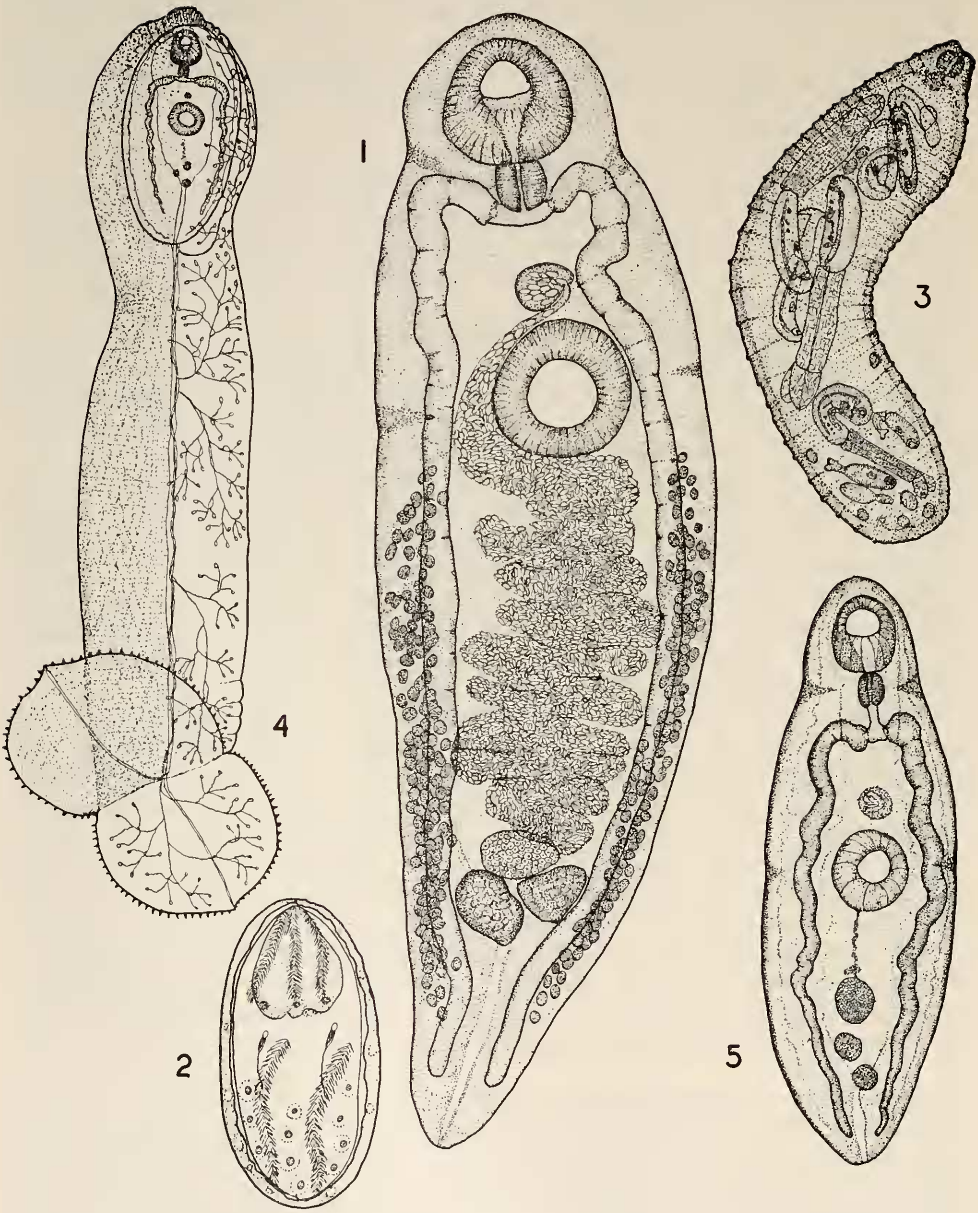

FIGURE 1. *Azygia acuminata,* adult (5.1 mm. in length), ventral view, collected from *Ameiurus nebulosus.*

FIGURE 2. *Azygia acuminata,* miracidium in egg (0.064 mm. in length), from sketches made of living larvae.

FIGURE 3. *Azygia acuminata,* redia (2.52 mm. in length), showing recognizable pharynx, annulations of the body and developing cercariae.

crawled out of the pharynx into the pharyngeal pocket. Up to six *A. sebago* larval distomes were seen to thus enter a single planarian. The larvae of *A. acuminata* were also enfolded by the planarians. The tail was sucked in but the larger size of the distome prevented both its entrance into the pharynx as well as into the aperture to the pharyngeal pocket. Occasionally a distome would be sucked part way into the pharynx and would remain so for several hours before finally becoming detached. Observations indicated that *D. tigrinum* could and probably does serve as a normal paratenic host for *A. sebago* but not for *A. acuminata*.

The cercaria of *A. acuminata* was found in *Campeloma decisum* obtained from the Santuit River, a small stream near the settlement of Santuit, Cape Cod, Mass. Small fishes, sticklebacks, *Eucalia inconstans* (Kirtland), *Fundulus heteroclitus,* the trout, *Salvelinus fontinalis* (Mitchill), and small eels, *Anguilla rostrata* LeSueur were obtained from streams known to be free of azygiid infections and were utilized as paratenic hosts. Specimens of each lot were examined and found to be free of azygiids and the remainder used, after infection, in feeding the definitive host fishes. These were found to be bull-heads, *Ameiurus nebulosus* LeSueur; blue gill sunfish, *Lepomis macrochirus* Raf.; chain pickerel, *Esox niger* LeSueur; and yellow perch, *Perca flavescens* (Mitchill). Yellow perch do not occur in this stream. These hosts were collected from a small pond known to be free of azygiids and were experimentally infected.

Large numbers of snails, *Campeloma decisum*, were kept in shallow, well aerated aquaria. In some instances liberated cercariae were pipetted from the aquarium and fed to small fish, and in other cases small fish were kept in the same aquarium. Heavy infections resulted in both cases since the swimming cercariae were readily ingested by the fish.

The young distomes differed in their relationship to the different species of small fish. Young trout and *Fundulus heteroclitus* retained an infection for only six to seven days. During this time the worms became less active and were found further and further posterior in the intestine of the fish. Sticklebacks and eels retained most of their infection in the stomach region. Worms removed from the latter species of fish were more active and their caeca were crowded with ingested material. Sticklebacks retained up to ten worms in the anterior portion of the intestine for as long as two weeks, whereas eels were found to have the stomach crowded with as many as twenty-four distomes.

Small fish were ingested by the larger fish used in the feeding experiments. The pectoral and dorsal spines of the sticklebacks were clipped before placing them in the aquarium with the definitive hosts.

Stages in the Life-Cycle

Adult (Fig. 1)

The original description of *A. acuminata* by Goldberger (1911) emphasized taxonomically-unimportant points such as zigzag caeca, rounded cephalic and

Figure 4. *Azygia acuminata* cercaria, naturally emerged, from sketches of living specimen (4.20 mm. in length). Flame cell pattern of the tail, transposed from sketches made from other cercariae, shown on right side of tail. Detail of musculature shown on the left, omitted on the right side of the tail.

Figure 5. *Azygia acuminata*, juvenile worm (1.18 mm. in length), experimental infection, from stomach of small eel, *Anguilla rostrata*.

pointed caudal ends, uninterrupted vitellaria and constricted neck region in his specific diagnosis.

Manter (1926) studied Goldberger's material, also material identified by A. R. Cooper, and specimens from H. B. Ward's collection. He suggested that certain similarities in the material showed *A. acuminata* to be a valid species. Manter stated (p. 61), "The most distinguishing specific characters were found to be: relatively wide body, anterior extent of the vitellaria, egg size, and poorly developed condition of the internal parenchyma muscles. It should be realized that the nature of all of these features is of somewhat precarious standing in this group. Probably no one of them, unless very marked, would justify recognition of a separate species. Only because of the general association of all of these characters can the forms be separated from the other common American species."

Van Cleave and Mueller (1934) regarded *A. acuminata* and *A. bulbosa* of Goldberger (1911) as synonyms, and since Manter had regarded *A. bulbosa* as a synonym of *A. longa,* they added *A. acuminata* to the long list of synonyms of *A. longa.* They pointed out that while the anterior extent of vitellaria was an important taxonomic character in the genus, the comparative width, constriction of the neck, and insignificant differences in egg size did not justify maintaining *A. acuminata* as a valid species. They stated that the less highly developed condition of the internal muscles was a dubious character. On the basis of the present study it appears evident that Van Cleave and Mueller were not justified in reducing *A. acuminata* to synonymy with *A. longa;* that *A. bulbosa* should be considered synonymous with *A. acuminata;* and the species should be maintained in the genus *Azygia.*

Emended description of the adult

Characters of the genus, body, 4–10 mm. long, width usually less than one-fifth of the length. Body characteristically slightly constricted anterior to the acetabulum. Position of constriction variable depending on state of contraction of the worm. Acetabulum one-fourth to one-third total length from anterior end of body. Genital pore and cirrus sac median in position immediately cephalad of acetabulum. Oral sucker 0.25–0.90 mm. in diameter, acetabulum slightly larger, 0.28–0.95 mm. in diameter. Gonads contiguous. Posterior testis larger than anterior testis. Vitellaria extra-caecal, extending from level of posterior margin of acetabulum to level midway between posterior testis and caudal end of body. Eggs consistently larger than in other described azygiid species, 0.64–0.69 × 0.30–0.34 mm.

Host: *Amia calva, Lepomis macrochirus,* and *Ameiurus nebulosus.*

Habitat: Stomach.

Localities: Illinois, Michigan, and Cape Cod, Massachusetts.

Miracidium

Miracidia representing all genera of the family Azygiidae have been described except for the genus *Leuceruthrus.* Azygiid miracidia lack cilia and are provided with bristle plates or plaques. The miracidium of *A. acuminata* is morphologically similar to the miracidium of *A. sebago* described by Stunkard (1956), to that of *Proterometra macrostoma* as reported by Hussey (1945), and to the earlier descriptions of that of *A. longa* by Schauinsland (1883) and Looss (1894).

Eggs must be ingested by the proper snail hosts before hatching will occur.

Attempts to hatch the eggs by placing them with material from the digestive tract of large *Campeloma decisum* were unsuccessful.

Miracidia were studied both alive in the egg (Fig. 2) and in stained sections of gravid worms. The miracidium almost completely fills the egg-shell. Radiating from the anterior end are five plaques bearing fine bristles arranged in chevron fashion with the apices anterior in position. These plaques extend posteriad about one-third of the length of the larva. A short distance from the posterior end of the miracidium, four other bristle plaques extend anteriad with a tendency to spiral, which may be due to movements of the larva.

Internally, occupying almost the anterior one-third of the miradicium, are four unicellular gland-like structures, the so-called primitive gut of earlier authors, which Stunkard considered probably secrete substances which aid in penetration. Wootton (1957) demonstrated that in *Allocreadium alloneotenicum* this group of glands does aid in penetration and that the penetration glands of earlier authors serve in forming the cuticle of the sporocyst. Attempts, in crushed snails, to observe the action of the anterior gland in the miracidium of *A. acuminata* were unsuccessful. It is possible since ciliary plates are lacking in azygiid miracidia that the miracidial covering serves also as the covering of the sporocyst, thus explaining the absence of cuticle-forming glands. Up to twelve germinal cells are visible in the posterior two-thirds of the miracidium. Paired flame cells lie near the middle of the body, one on each side, each with a duct leading caudad. The ducts could not be clearly traced to their pores.

Sporocyst

Young snails were removed directly from the uterus of an uninfected female *Campeloma decisum*. The young snails readily fed in a layer of clean sand. Eggs of *A. acuminata*, each containing an active miracidium, were added to the sand in which the snails were feeding. Snails were dissected at the end of one, two, and three weeks but no infection was found. It is probable that the snails must be larger before infection will occur.

Rediae

Infections with larvae of *A. acuminata* were present only in females of *Campeloma decisum*. Infections of male *Campeloma* were never observed. The redial stages were present, usually with unencysted metacercariae identified as *Leucochloridiomorpha constantiae* (Mueller), in the uterus of the snail. The loci of infection did not extend into the digestive gland, the usual site of infection for larval trematodes. Up to eighty rediae were dissected from the uterus of a single snail. These, plus almost as many metacercariae of *L. constantiae*, completely filled the uterus. Embryo snails, which were usually found in the uterus of uninfected females, were absent in infected snails. In a few instances partially empty embryo snail shells were found in the uterus, but it was evident that infection with larval *A. acuminata* adversely affected normal development of the young snails. The redial stages undoubtedly derived their nourishment from the developing embryo snails within the uterus. Young snails develop normally when associated only with metacercariae of *L. constantiae*. Thus, infection of the snail with larvae of

A. acuminata caused the degeneration of the young snails. Rediae (Fig. 3) when fixed averaged 2.15 × 0.71 mm.

The vermiform rediae are very active and are capable of extending to a length of over 5 mm. When contracted, the external body wall is formed into regularly spaced annular rings, giving it a wrinkled appearance. Even when fully extended, these rings persist as fine annular projections. The body wall is 0.043 mm. in thickness in sectioned material. The small pharynx is not easily observable in stained whole mounts, but is visible in sections as a rudimentary structure (0.11 mm. in diameter) with a well defined lumen. No recognizable birth pore can be seen, either in sectioned materials or in stained whole mounts or rediae. It is probable that cercariae escape through the pharynx.

Up to twelve recognizable cercariae are present in each redia, with seven to ten being the usual number. Other developing cercariae are present as germ-balls, and a maximum number of twelve developing cercariae present with six germ-balls was observed. The redial stage is very similar to that reported by Szidat (1932) for *A. lucii* and by Stunkard (1956) for *A. sebago,* varying only in size and in number of developing cercariae.

Cercariae

The development of the cercaria is typical of that reported for cystocercous larvae. The tail becomes evident in development when the larva reaches a length of only about 0.1 mm. When the larva reaches a total length of 0.5–0.6 mm. the furcal buds appear as oval projections. At this stage the suckers are recognizable, and the primordia of one testis and the cirrus sac are also visible as deeper staining areas. The tail increases more rapidly than the distome during further development. The largest cercaria observed within a redia as a stained whole mount measured 2.22 mm. in total length. The distome measured 0.74 × 0.37 mm.

Cercariae appear to undergo additional development in the uterus of the snail before escaping from the female genital pore. After emergence, they are quiescent for a brief period before actively swimming in a typical cystocercous fashion. Cercariae normally emerge between 12:00 P.M. and 4:00 A.M. Standard Time. Limited numbers escape during daylight hours. It appears, however, that the majority emerge during hours of darkness and are either ingested by small fish at this time or in the early hours of daylight. They live for only ten to twelve hours, becoming less active as they age. Cercariae while still within the uterus of the snail do not have the distome portion enclosed by the tail. Upon coming into contact with the water, the anterior tail bulb absorbs water, expands rapidly anteriorly thus enclosing the distome, as has been described for other cyctocercous cercariae.

Mature, normally liberated cercariae (Fig. 4) measure 3.21–4.69 mm. (averaged 4.22 mm.) in total length when infected snails are first brought into the laboratory. The size of the cercariae gradually decreases in snails kept in captivity, undoubtedly a result of deficient nutrition of the hosts. The tail stem is round in cross-section at the bulb-like anterior end enclosing the distome. This portion measures 0.69–0.96 mm. in diameter (average 0.78 mm.). Just posterior to the more or less rigid anterior bulb enclosing the distome, the tail decreases slightly to a diameter of 0.52–0.82 mm. (average 0.66 mm.). From the constricted neck-portion the tail gradually flattens and widens to a width of 0.62–1.11 mm. (average 0.84 mm.) and then tapers gradually to an average width of 0.79 mm. just anterior to the furcal branches.

The furci are broadly lobed structures, 1.11–1.28 mm. in length (average 1.21 mm.) and 0.89–1.09 mm. in width (average 0.94 mm.). Each furca has a terminal papilla on which the excretory pore opens and small regularly arranged scale-like marginal protuberances. The tail of the cercaria is colorless, slightly opaque and devoid of protuberances, spines and mammulations characteristic of *C. mirabilis* Braun, 1891, *C. macrostoma* Faust, 1918, *C. splendens* Szidat, 1932, *C. anchoroides* Ward, 1916, and *C. sebago* Stunkard, 1956. The cercaria of *A. acuminata* differs in size and in the proportionate size of the distome when compared with the tail length from other cystocercous cercariae which characteristically do not possess papillae. It differs from *A. hodgesiana* Smith, 1932 since the genital organs are not functional as they are in the latter; from *A. stephanocauda* Faust, 1921 in size and shape of the tail; from *C. wrighti* Ward, 1916 in size; and from *C. pekinensis* Faust, 1921 in proportionate size of the distome.

The cercaria of *A. acuminata* is most like *C. brookoveri* Faust, 1918 and *C. anchoroides* Ward, 1916, but is over twice as large. *C. brookoveri* was originally described from crushed *Campeloma* sp. and the free-swimming larva was rediscovered by Dickerman (1937) from the same snails. Unfortunately Dickerman did not further describe the species. *C. anchoroides* was collected only in plankton tows from Lake Erie. The size and obvious similarities in structure of the two forms, as well as geographic proximity of the type localities, caused Horsfall (1934) to think that they will prove to be synonymous when the life-cycles are known.

The enclosed distome measures 0.66–0.79 × 0.37–0.47 mm. in living material. It usually lies with the oral sucker at the anterior end of the tail-bulb. It is flattened and varies as to its orientation to the width of the tail, sometimes being at right angles and at others with its width the same as the width of the tail. The excretory system is continuous with that of the tail, extending down the tail as a common excretory canal bifurcating at the bases of the furci and opening at the small points of the furci.

The structure of the larva when forced from the tail-bulb is typically azygiid. The preacetabular region bears many papillae which decrease in size and number, and are absent behind the mid-acetabular region. Living specimens, flattened slightly under a cover glass, measure from 0.98–1.38 mm. (average 1.18 mm.) in length and from 0.37–0.54 mm. in width (average 0.48 mm.). The oral sucker varies from 0.22–0.25 mm. in length and from 0.20–0.23 mm. in width. It is subterminal, opening ventrally. The pharynx measures 0.090–0.098 mm. in length and 0.49–0.61 mm. in width. The acetabulum varies from 0.22–0.25 mm. in length and from 0.25–0.29 mm. in width. It is about midlength in the body. The digestive caeca are filled with opaque material and extend almost to the posterior end of the larva. The excretory bladder extends anteriorly to the region just posterior to the testes where it branches into two main collecting ducts. These extend median to the caeca, crossing laterad as the caeca turn mediad to join the pharynx. After crossing under the caeca, the ducts pass laterally and antero-laterally to the oral sucker, continuing almost to the anterior end of the body, but they do not join. Anterio-lateral to the oral sucker each duct doubles backward and extends posteriad, lateral to the caeca, giving off eleven branches.

The first branch is located lateral to the oral sucker, the second at the level of the pharynx, the third and fourth anterior to the acetabulum, the fifth at the anterior edge of the acetabulum, the sixth lateral to the acetabulum, the seventh and eighth are close together just behind the acetabulum, the ninth and tenth are about equally spaced in the intervening region, while the last branch continues to the posterior end of the body lateral to the excretory bladder. Each branch divides three times in a dichotomous fashion, thus forming two primary, four secondary, and eight tertiary branches. Each tertiary branch drains four flame cells. Thus the flame cell formula is 2 (11 × 32) or 704 flame cells.

The number and arrangement of the flame cells is in agreement with those reported by Looss (1894) for *Azygia terreticolla* (= *A. lucii*) and by Stunkard (1956) for *A. sebago*. While the numbers of branches and flame cells agree with these earlier descriptions, the positions of the branches are different in *A. acuminata* due to the relatively more posterior position of the acetabulum.

The excretory system of the tail is equally complex (Fig. 4). In addition to the common excretory canal extending down the center of the tail and bifurcating into each furca, two paired accessory canals paralleling the main canal were observed. One duct and its branches drained the right side of the tail and the right furca, and the other the left side and the left furca. Each duct collected from five branches but dichotomous bifurcation of the ducts was not as clearly evident as in the distome portion.

Each of the five branches, however, did drain from 32 flame cells, arranged in groups of fours. The first branch turned anteriad from just caudad of the distome, draining the enclosing bulb area, the second and third branches joined the collecting duct close together in about the middle of the tail, the second turning anteriad and draining that area, the third posteriad collecting from the third quarter of the tail. The fourth branch joined the collecting ducts about three-fourths the length of the tail and drained the final quarter of the tail. From the fourth branch, the duct extended into a furca draining from 32 flame cells. The formula for the tail is thus 2 (5 × 32) or 320 and the entire cercaria has a formula of 2 (16 × 32) or 1,024 flame cells.

The connection of the accessory ducts of the tail to the rest of the excretory system was not resolved. The dense protoplasm at the tip of the tail in immature cercariae freed from rediae and the congested area at the base of distomes enclosed in the tail of normally liberated cercariae made observations impossible. These ducts are 0.011 mm. in diameter compared to the common duct which is 0.055 mm. in diameter. Faust (1921) reported that in *C. pekinensis*, the tail had only 32 flame cells and connected to the excretory system of the distome as the eighth branch. He further reported only seven branches in the distome portion of *C. pekinensis*. The flame cell pattern of this form should be examined in the light of the observations of the cercariae of *A. lucii* Looss (1894), *A. sebago* Stunkard 1956, and the present observations of *A. acuminata*, since *C. pekinensis* would appear to also develop into an azygiid.

Young worms

Worms increased very little in size and did not undergo further development while in the stomach of small fishes. A young distome (Fig. 5) from the stomach of a small eel differed from one newly forced from the cercarial tail only in the size of the caeca. In worms taken from paratenic hosts, the caeca were enlarged with food particles. No measureable differences in worms from various hosts were found. Worms from the sticklebacks and young eels were more active and appeared healthier than worms from other small fish.

Manter's synopsis and key to the genus *Azygia* can be revised to include *A. sebago* and the cercaria of each species can be noted as follows:

KEY TO THE SPECIES OF AZYGIA FROM NORTH AMERICA

1 (2) Vitellaria not extending appreciably posterior to the last testis. Length 6–54 mm. (*C. mirabilis* Braun, 1891) ....*A. lucii* (Mueller)
2 (1) Vitellaria extending posteriad at least half the distance between posterior testis and end of body 3
3 (4) Acetabulum near middle of body, gonads in posterior one-sixth of body (*C. angusticauda* Dickerman, 1937) ....*A. angusticauda* (Stafford, 1904)
4 (3) Acetabulum within anterior one-third of body, gonads more anterior 5
5 (6) Body width usually one-fifth the total length, vitellaria extending posteriad from close behind acetabulum, internal parenchyma muscles relatively weak. Eggs 0.064–0.069 × 0.30–0.34 mm. (*Cercaria acuminata,* present paper) .....*A. acuminata* Goldberger, 1911
6 (7) Body width proportionately less than one-fifth the length, vitellaria begin some distance posterior to acetabulum, internal parenchyma muscles strongly developed, eggs variable in size but smaller than *A. acuminata* 7
7 (8) Body length not over 15 mm., usually smaller, body robust in appearance (*Cercaria sebago* Stunkard, 1956) ....*A. sebago* Ward, 1910
8 (7) Body often extremely elongate, vitellaria beginning proportionally more posteriorly (*Cercaria longa* Sillman, 1953a) ....*A. longa* (Leidy, 1851).

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Summary

1. Stages in the life-cycle of *Azygia acuminata* are described and figured. Cystocercous cercariae develop from rediae in the snail, *Campeloma decisum.* The cercaria is morphologically distinct from other described cystocercous cercariae. Rediae are similar to the same stage described for other members of the genus, but are unique since they develop in the uterus of female *Campeloma decisum.*

2. The excretory system of the cercaria is complex, showing a formula of 2 (11 × 32) or 704 flame cells for the distome portion and 2 (5 × 32) or 320 flame cells in the tail. The excretory formula of the cercariae is thus 2 (16 × 32) or 1,024 flame cells.

3. Attempts to experimentally infect small snails, taken from the uterus of a *Campeloma decisum,* by feeding them eggs of *A. acuminata* were not successful.

4. Various small fishes were utilized as paratenic hosts by the young distomes. Infections in sticklebacks, *Eucalia inconstans,* and small eels, *Anguilla rostrata,* resulted in more active and vigorous worms than did infections from other paratenic hosts.

5. The variation that normally occurs in members of the genus *Azygia* due to development in a wide variety of hosts is not known. Consequently diagnostic characters of mature worms can not be relied on to distinguish species. On the basis of this report the suppressed species *A. acuminata* is regarded as a valid species and should be retained in the genus *Azygia.*

6. *A. acuminata,* previously reported only from *Amia calva,* was found occur-

ring naturally in bullheads, *Ameiurus nebulosus,* blue gill sunfish, *Lepomis macrochirus,* and chain pickerel, *Esox niger,* from Santuit River, Barnstable County, Cape Cod, Massachussetts. Experimental infections were also obtained in these fishes and in the yellow perch, *Perca flavescens.*

7. A revised key for the genus *Azygia* is presented, listing the recognized species and the described cercarial stages.

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